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=> d his

     (FILE 'HOME' ENTERED AT 15:47:50 ON 03 MAR 2004)

     FILE 'CAPLUS' ENTERED AT 15:48:29 ON 03 MAR 2004
L1          2184 S (RNA (2W)DEPENDENT) (W)(RNA (W) POLYMERASE)
L2             8 S L1 AND INTERFACE

     FILE 'USPATFULL' ENTERED AT 15:51:12 ON 03 MAR 2004
L3            43 S L1 AND INTERFACE
L4            77 S ((RNA (2W)DEPENDENT) (W)(RNA (W) POLYMERASE))/TI,CLM,AB
L5            62 S L4 AND (INHIBIT? OR INTERACT? OR PHARMACOPHORE)
L6             6 S L5 AND INTERFACE
L7            12 S L5 AND 342

     FILE 'REGISTRY' ENTERED AT 16:11:42 ON 03 MAR 2004
L8             0 S KPHKCTFEGCRKSYSRLENLLKTHLRSH/SQSP
L9            25 S KPHKCTFEGCRKSYSRLENLKTHLRSH/SQSP

     FILE 'CAPLUS' ENTERED AT 16:18:28 ON 03 MAR 2004
L10           11 S L9
L11            0 S (RNA (W) POLYMERASE) AND L9
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(FILE 'HOME' ENTERED AT 15:47:50 ON 03 MAR 2004)

FILE 'CAPLUS' ENTERED AT 15:48:29 ON 03 MAR 2004
L1 2184 S (RNA (2W)DEPENDENT) (W)(RNA (W) POLYMERASE)
L2 8 S L1 AND INTERFACE

=> d bib, abs 1-8

L2 ANSWER 1 OF 8 CAPLUS COPYRIGHT 2004 ACS on STN
AN 2003:778896 CAPLUS
DN 140:138497
TI Targeting NS5B **RNA-dependent RNA polymerase** for anti-HCV chemotherapy
AU Wu, Jim Zhen; Hong, Zhi
CS Drug Discovery, Ribapharm, Inc., Costa Mesa, CA, 92626, USA
SO Current Drug Targets: Infectious Disorders (2003), 3(3), 207-219
CODEN: CDTIAS; ISSN: 1568-0053
PB Bentham Science Publishers Ltd.
DT Journal; General Review
LA English
AB A review. The global prevalence of persistent hepatitis C virus (HCV) infection and the lack of a highly effective and well-tolerated antiviral therapy have spurred intensive efforts to discover and develop novel anti-HCV therapy in the pharmaceutical industry. HCV NS5B **RNA-dependent RNA polymerase** (RdRp), the centerpiece for viral replication, constitutes a valid target for drug discovery. Compared to the host RNA and DNA polymerases, NS5B RdRp has distinct subcellular localization at the **interface** of the endoplasmic reticulum (ER) membrane and cytoplasm, a novel catalytic mechanism and many unique structural features, all of which make it an attractive target for developing effective anti-HCV therapeutics. High genetic variation among the major HCV genotypes commands that any efficacious NS5B inhibitors have to be broadly active against NS5Bs from various genotypes. Rapid viral replication and its inherent genetic diversity will certainly culminate drug resistance to any NS5B inhibitors. Therefore, iterative drug design and combination therapies of drugs that intervene with different steps in the HCV replicative cycle are needed to combat the viral infection. Many classes of nucleoside and non-nucleoside inhibitors of NS5B RdRp have been identified and appeared in literatures and patent applications. This progress holds considerable promise to the development of novel, specific and highly effective therapeutics to achieve sustained response and ultimately the eradication of HCV infection.
RE.CNT 74 THERE ARE 74 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L2 ANSWER 2 OF 8 CAPLUS COPYRIGHT 2004 ACS on STN
AN 2003:189124 CAPLUS
DN 139:18989
TI Non-nucleoside Analogue Inhibitors Bind to an Allosteric Site on HCV NS5B Polymerase
AU Wang, Meitian; Ng, Kenneth K.-S.; Cherney, Maia M.; Chan, Laval; Yannopoulos, Constantin G.; Bedard, Jean; Morin, Nicolas; Nguyen-Ba, Nghe; Alaoui-Ismaili, Moulay H.; Bethell, Richard C.; James, Michael N. G.
CS Department of Biochemistry, Canadian Institutes for Health Research Group in Protein Structure and Function, University of Alberta, Edmonton, AB, T6G 2H7, Can.
SO Journal of Biological Chemistry (2003), 278(11), 9489-9495
CODEN: JBCHA3; ISSN: 0021-9258
PB American Society for Biochemistry and Molecular Biology
DT Journal
LA English
AB X-ray crystal structures of two non-nucleoside analog inhibitors bound to hepatitis C virus NS5B **RNA-dependent RNA**

polymerase have been determined to 2.0 and 2.9 Å resolution These noncompetitive inhibitors bind to the same site on the protein, .apprx.35 Å from the active site. The common features of binding include a large hydrophobic region and two hydrogen bonds between both oxygen atoms of a carboxylate group on the inhibitor and two main chain amide nitrogen atoms of Ser476 and Tyr477 on NS5B. The inhibitor-binding site lies at the base of the thumb domain, near its **interface** with the C-terminal extension of NS5B. The location of this inhibitor-binding site suggests that the binding of these inhibitors interferes with a conformational change essential for the activity of the polymerase.

RE.CNT 48 THERE ARE 48 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L2 ANSWER 3 OF 8 CAPLUS COPYRIGHT 2004 ACS on STN
AN 2002:838645 CAPLUS
DN 138:85370
TI Poliovirus **RNA-dependent RNA** **polymerase** (3Dpol): structure, function, and mechanism
AU Cameron, Craig E.; Gohara, David W.; Arnold, Jamie J.
CS Department of Biochemistry and Molecular Biology, Pennsylvania State University, University Park, PA, 16802, USA
SO Molecular Biology of Picornaviruses (2002), 255-267. Editor(s): Semler, Bert L.; Wimmer, Eckard. Publisher: American Society for Microbiology, Washington, D. C.
CODEN: 69DFWL; ISBN: 1-55581-210-4
DT Conference; General Review
LA English
AB A review. In this chapter, we have shown that over the past few years a remarkable amount of information has become available that has increased our understanding of the structure, function, and mechanism of 3Dpol. In fact, in most respects, our understanding of 3Dpol is now equivalent to that of other classes of nucleic acid polymerase. Clearly, much work remains to be completed. A few addnl. structures will have a significant impact on our understanding of the structure-function relationships of 3Dpol. For example, a complete structure of 3Dpol is needed to solve the mystery surrounding **interface** II. A structure of 3Dpol in complex with substrates is needed to define the path of the nucleic acid through the enzyme, thus providing a structural basis for the increased affinity for nascent RNA relative to template. Given the many interactions between 3Dpol and other viral nonstructural proteins and cellular proteins, a more quant. anal. of these interactions may shed light on replicase composition, stoichiometry, and assembly, thus facilitating reconstitution of genome replication in vitro from purified components. Finally, while a variety of assays exist now that permit elucidation of the biochem. and biophys. properties of 3Dpol, it is important to begin to interrogate more rigorously the biol. relevance of these properties.

RE.CNT 101 THERE ARE 101 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L2 ANSWER 4 OF 8 CAPLUS COPYRIGHT 2004 ACS on STN
AN 2002:677939 CAPLUS
DN 137:365434
TI Structure-function relationships of the **RNA-dependent** **RNA polymerase** from poliovirus (3Dpol). A surface of the primary oligomerization domain functions in capsid precursor processing and VPg uridylylation
AU Pathak, Harsh B.; Ghosh, Saikat Kumar B.; Roberts, Allan W.; Sharma, Suresh D.; Yoder, Joshua D.; Arnold, Jamie J.; Gohara, David W.; Barton, David J.; Paul, Aniko V.; Cameron, Craig E.
CS Department of Biochemistry and Molecular Biology, Pennsylvania State University, University Park, PA, 16802, USA
SO Journal of Biological Chemistry (2002), 277(35), 31551-31562
CODEN: JBCHA3; ISSN: 0021-9258
PB American Society for Biochemistry and Molecular Biology

DT Journal
LA English
AB The primary oligomerization domain of poliovirus polymerase, 3Dpol, is stabilized by the interaction of the back of the thumb subdomain of one mol. with the back of the palm subdomain of a second mol., thus permitting the head-to-tail assembly of 3Dpol monomers into long fibers. The interaction of Arg-455 and Arg-456 of the thumb with Asp-339, Ser-341, and Asp-349 of the palm is key to the stability of this **interface**. We show that mutations predicted to completely disrupt this **interface** do not produce equivalent growth phenotypes. Virus encoding a polymerase with changes of both residues of the thumb to alanine is not viable; however, virus encoding a polymerase with changes of all three residues of the palm to alanine is viable. Biochem. anal. of 3Dpol derivs. containing the thumb or palm substitutions revealed that these derivs. are both incapable of forming long fibers, suggesting that polymerase fibers are not essential for virus viability. The RNA binding activity, polymerase activity, and thermal stability of these derivs. were equivalent to that of the wild-type enzyme. The two significant differences observed for the thumb mutant were a modest reduction in the ability of the altered 3CD proteinase to process the VP0/VP3 capsid precursor and a substantial reduction in the ability of the altered 3Dpol to catalyze oriI-templated uridylylation of VPg. The defect to uridylylation was a result of the inability of 3CD to stimulate this reaction. Because 3C alone can substitute for 3CD in this reaction, we conclude that the lethal replication phenotype associated with the thumb mutant is caused, in part, by the disruption of an interaction between the back of the thumb of 3Dpol and some undefined domain of 3C. We speculate that this interaction may also be critical for assembly of other complexes required for poliovirus genome replication.
RE.CNT 39 THERE ARE 39 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L2 ANSWER 5 OF 8 CAPLUS COPYRIGHT 2004 ACS on STN
AN 2002:248515 CAPLUS
DN 136:365660
TI Oligomerization and cooperative RNA synthesis activity of hepatitis C virus **RNA-dependent RNA polymerase**
AU Wang, Q. May; Hockman, Michelle A.; Staschke, Kirk; Johnson, Robert B.; Case, Katharine A.; Lu, Jirong; Parsons, Steve; Zhang, Faming; Rathnachalam, Radhakrishnan; Kirkegaard, Karla; Colacino, Joseph M.
CS Lilly Research Laboratories, Eli Lilly and Company, Indianapolis, IN, 46285, USA
SO Journal of Virology (2002), 76(8), 3865-3872
CODEN: JOVIAM; ISSN: 0022-538X
PB American Society for Microbiology
DT Journal
LA English
AB The NS5B **RNA-dependent RNA polymerase** encoded by hepatitis C virus (HCV) plays a key role in viral replication. Reported here is evidence that HCV NS5B polymerase acts as a functional oligomer. Oligomerization of HCV NS5B protein was demonstrated by gel filtration, chemical crosslinking, temperature sensitivity, and yeast cell two-hybrid anal. Mutagenesis studies showed that the C-terminal hydrophobic region of the protein was not essential for its oligomerization. Importantly, HCV NS5B polymerase exhibited cooperative RNA synthesis activity with a dissociation constant, Kd, of ≈22 nM, suggesting a role for the polymerase-polymerase interaction in the regulation of HCV replicase activity. Further functional evidence includes the inhibition of the wild-type NS5B polymerase activity by a catalytically inactive form of NS5B. Finally, the X-ray crystal structure of HCV NS5B polymerase was solved at 2.9 Å. Two extensive **interfaces** have been identified from the packing of the NS5B mols. in the crystal lattice, suggesting a higher-order structure that is

consistent with the biochem. data.
RE.CNT 42 THERE ARE 42 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L2 ANSWER 6 OF 8 CAPLUS COPYRIGHT 2004 ACS on STN
AN 2001:868251 CAPLUS
DN 136:15221
TI Viral **RNA-dependent RNA polymerase** pharmacophores for inhibitors of viral infection
IN Kirkegaard, Karla A.; Richmond, Kathy; Lyle, John M.; Schulz, Steve; Hobson, Scott
PA The Board of Trustees of the Leland Stanford Junior University, USA
SO PCT Int. Appl., 52 pp.
CODEN: PIXXD2
DT Patent
LA English
FAN.CNT 1

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI WO 2001089560	A1	20011129	WO 2001-US16674	20010522
W: AU, CA, JP				
RW: AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR				
US 2002164348	A1	20021107	US 2001-863841	20010522
PRAI US 2000-206887P	P	20000524		

AB The invention provides a viral **RNA-dependent RNA polymerase** pharmacophore which is characterized by binding to a conserved **interface** binding surfaces of a viral **RNA-dependent RNA polymerase**.
RE.CNT 1 THERE ARE 1 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L2 ANSWER 7 OF 8 CAPLUS COPYRIGHT 2004 ACS on STN
AN 2001:613078 CAPLUS
DN 135:329013
TI Functional analysis of PA binding by influenza A virus PB1: effects on polymerase activity and viral infectivity
AU Perez, Daniel R.; Donis, Ruben O.
CS Department of Veterinary and Biomedical Sciences, University of Nebraska-Lincoln, Lincoln, NE, 68583-0905, USA
SO Journal of Virology (2001), 75(17), 8127-8136
CODEN: JOVIAM; ISSN: 0022-538X
PB American Society for Microbiology
DT Journal
LA English
AB Influenza A virus expresses three viral polymerase (P) subunits-PB1, PB2, and PA-all of which are essential for RNA and viral replication. The functions of P proteins in transcription and replication have been partially elucidated, yet some of these functions seem to be dependent on the formation of a heterotrimer for optimal viral RNA transcription and replication. Although it is conceivable that heterotrimer subunit interactions may allow a more efficient catalysis, direct evidence of their essentiality for viral replication is lacking. Biochem. studies addressing the mol. anatomy of the P complexes have revealed direct interactions between PB1 and PB2 as well as between PB1 and PA. Previous studies have shown that the N-terminal 48 amino acids of PB1, termed domain α, contain the residues required for binding PA. We report here the refined mapping of the amino acid sequences within this small region of PB1 that are indispensable for binding PA by deletion mutagenesis of PB1 in a two-hybrid assay. Subsequently, we used site-directed mutagenesis to identify the critical amino acid residues of PB1 for interaction with PA in vivo. The first 12 amino acids of PB1 were found to constitute the core of the interaction **interface**, thus narrowing the previous boundaries of domain α. The role of the

minimal PB1 domain α in influenza virus gene expression and genome replication was subsequently analyzed by evaluating the activity of a set of PB1 mutants in a model reporter minigenome system. A strong correlation was observed between a functional PA binding site on PB1 and P activity. Influenza viruses bearing mutant PB1 genes were recovered using a plasmid-based influenza virus reverse genetics system. Interestingly, mutations that rendered PB1 unable to bind PA were either nonviable or severely growth impaired. These data are consistent with an essential role for the N terminus of PB1 in binding PA, P activity, and virus growth.

RE.CNT 45 THERE ARE 45 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L2 ANSWER 8 OF 8 CAPLUS COPYRIGHT 2004 ACS on STN
AN 1997:744763 CAPLUS
DN 128:32259
TI Genetic dissection of interaction between poliovirus 3D polymerase and viral protein 3AB
AU Hope, Debra A.; Diamond, Scott E.; Kirkegaard, Karla
CS Dep. of Mol., Cell. and Dev. Biol., Howard Hughes Med. Inst., Univ. of Colorado, Boulder, CO, 80309, USA
SO Journal of Virology (1997), 71(12), 9490-9498
CODEN: JOVIAM; ISSN: 0022-538X
PB American Society for Microbiology
DT Journal
LA English
AB Poliovirus **RNA-dependent RNA polymerase** 3D and viral protein 3AB are both thought to be required for the initiation of RNA synthesis. These two proteins phys. associate with each other and with viral RNA replication complexes found on virus-induced membranes in infected cells. An understanding of the **interface** between 3D and 3AB would provide a first step in visualizing the architecture of the multiprotein complex that is assembled during poliovirus infection to replicate and package the viral RNA genome. The identification of mutations in 3D that diminish 3D-3AB interactions without affecting other functions of 3D polymerase is needed to study the function of the 3D-3AB interaction in infected cells. We describe the use of the yeast two-hybrid system to isolate and characterize mutations in 3D polymerase that cause it to interact less efficiently with 3AB than wild-type polymerase. One mutation, a substitution of leucine for valine at position 391 (V391L), resulted in a 3AB-specific interaction defect in the two-hybrid system, causing a reduction in the interaction of 3D polymerase with 3AB but not with another viral protein or a host protein tested. In vitro, purified 3D-V391L polymerase bound to membrane-associated 3AB with reduced affinity. Poliovirus that contained the 3D-V391L mutation was temperature sensitive, displaying a pronounced conditional defect in RNA synthesis. We conclude that interaction between 3AB and 3D or 3D-containing polypeptides plays a role in RNA synthesis during poliovirus infection.

RE.CNT 38 THERE ARE 38 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT